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IN THE UNITED STATES PATENT & TRADEMARK OFFICE

IN RE APPLICATION OF :
BERNARD PAU, ET AL. : EXAMINER: AEDER, SEAN E
SERIAL NO: 10/516,946 :
FILED: AUGUST 9, 2005 : GROUP ART UNIT: 1642
FOR: OXALIPLATIN ANTI- :
RESISTANCE AGENT

APPEAL BRIEF

COMMISSIONER FOR PATENTS
ALEXANDRIA, VIRGINIA 22313

SIR:

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(i) Real Party in Interest

Centre National de La Recherche Scientifique (CNRS), Université de Montpellier I,
and Institut Pasteur are the real parties in interest.

(ii) Related Appeals or Interferences

The Appellants are unaware of any related appeals or interferences that would directly affect, be directly affected by, or have a bearing on the Board's decision in this appeal.

(iii) Status of the Claims

Claims 1, 2, 5, 8, 10-12, 24 and 27 are on Appeal. Claims 1 and 27 are the only independent claims on Appeal.

Claims 6, 7, 9, 13, and 19-23 have been withdrawn from consideration as being directed to a non-elected invention.

Claims 3, 4, 14-18, 25 and 26 have been cancelled.

The Claims Appendix below provides a clean copy of the claims on appeal.

(iv) Status of the Amendment

The Amendment filed on October 5, 2009 after final rejection has been entered for purposes of Appeal (see the Advisory Action dated October 13, 2009).

(v) Summary of the Claimed Subject Matter

The invention pertains to a process for determining whether a cancer cell is resistant to a broad-spectrum anti-tumor agent called oxaliplatin. Oxaliplatin “is a platin salt possessing an anti-tumor activity spectrum much broader than conventional platin salts such as cisplatin or carboplatin” (specification, page 2, lines 22-24).

By determining whether a cancer or cancer cell is resistant to oxapliplatin a doctor can avoid subjecting a patient to the toxicity and risk of a chemotherapy that would have no therapeutic benefit and can choose instead medications that can circumvent or oppose the resistance mechanisms (specification, page 2, lines 15-19).

The mechanisms associated with the resistance of cancer cells to oxaliplatin “remained unexplained until this invention” (specification, page 2, line 31). The inventors have shown that oxaliplatin resistance “is associated with abnormal expression of the mitochondrial apoptosis genes” (specification, page 4, lines 27-29), such as the gene expressing Bax and/or Bak proteins (specification, page 13, lines 10-12, Examples 1-2 on page 27, line 30-page 36, line 6).

It was discovered that “the state of resistance or sensitivity of cells to oxaliplatin correlates well with the degree of activation of Bax” (specification, page 34, lines 22-24) and that as “in the case with Bax, activation of Bak correlates well with the response of the cells to oxaliplatin” (specification, page 35, last line-page 36). “Early activation of Bax and Bak is associated with the state of sensitivity of the cells to oxaliplatin” (specification, page 36, last sentence).

Support for independent claims 1 and 19 is annotated within **embolded** brackets below:

Claim 1: A process for detecting resistance of a cancer cell to oxaliplatin treatment
[specification, page 1, lines 3-7, original claims] comprising:

detecting the expression of an effector or marker gene expressing the pro-apoptotic Bax and/or Bak protein(s) [specification, page 6, lines 14-15; Examples 1-2] in a cancer cell and in a control cell not resistant to oxaliplatin;

comparing said expression between said cancer cell and said control cell [specification, page 8, last paragraph], wherein reduced expression of said effector or marker gene in said cancer cell compared to said control cell indicates that said cancer cell is resistant to oxaliplatin [specification, page 9, first paragraph; original claim 13, step b].

Claim 27: A process for detecting resistance of a cancer cell to oxaliplatin treatment [specification, page 1, lines 3-7, original claims] comprising:

measuring the level of expression of the mRNA(s) [specification, page 8, lines 21-22; original claim 5] encoding the pro-apoptotic Bax and/or Bak protein(s) [Examples 1-2] in a cancer cell and in a control cell not resistant to oxaliplatin;

comparing said expression between the cancer cell and the control cell [specification, page 8, last paragraph],

wherein a lower level of expression of said mRNA in the cancer cell compared to a control cell not resistant to oxaliplatin is indicative of resistance to oxaliplatin [specification, page 9, first paragraph; original claim 13, step b].

(vi) Grounds of Rejection to be Reviewed on Appeal

A. Whether claims 1, 2, 5, 8, 10, 12, 24 and 27 are unpatentable under 35 U.S.C. §103(a) as being obvious over Maurer, et al., Dig. Dis. Sci. 43:2641, in view of Macpherson, et al., PAACR Ann. Mtg. 43:407 and Chao, et al., J. Exp. Med. 182:821.

B. Whether claims 1, 2, 5, 8, 10-12, 24 and 27 are unpatentable under 35 U.S.C. §103(a) as being obvious over Maurer, et al., Dig. Dis. Sci. 43:2641, in view of Macpherson, et al., PAACR Ann. Mtg. 43:407 and Chao, et al., J. Exp. Med. 182:821, and further in view of Aggarwal, et al., J. Immunol. 160:1627.

(vii) Arguments

Issue A: Rejection—35 U.S.C. §103

Claims 1, 2, 5, 8, 10, 12, 24 and 27 stand rejected under 35 U.S.C. §103(a) as being unpatentable over Maurer, et al., Dig. Dis. Sci. 43:2641, in view of Macpherson, et al., PAACR Ann. Mtg. 43:407 and Chao, et al., J. Exp. Med. 182:821. This rejection cannot be sustained because none of these references suggests that a reduced level of expression of Bax gene alone correlates with enhanced resistance to oxaliplatin. Consequently, the prior art does not disclose or suggest all the elements of the invention and the Office has not established a *prima facie* case for obviousness. Moreover, the Examiner has not clearly articulated reasons why the invention would have been obvious to one of ordinary skill in the art at the time of invention.

Maurer did not disclose all the method steps of the present invention, specifically the comparisons between gene expression of pro-apoptotic Bax and/or Bak proteins in cancer cells and gene expression in control cells not resistant to oxaliplatin.

The secondary references Macpherson and Chao do not concern detecting the differences between Bax and Bcl expression in tumor and control cells and fail to disclose these elements of the invention as well. Neither of these documents suggests that oxaliplatin resistance correlates with Bax gene expression alone.

Macpherson discloses that the down-regulation of Bcl-xl (knock-out mediated by prior antisense) enhances the apoptotic response to oxaliplatin and consequently enhances oxaliplatin cytotoxicity. However, this document did not suggest comparing the level of expression of the gene encoding Bcl-xl in tumor and control cells to determine the degree of oxaliplatin resistance.

In distinction to determining the relative level of expression of Bax and/or Bak between cancer and control cells, Chao teaches an entirely different parameter: the ratio of **heterodimerization** of Bcl-2 and Bax as shown by the citations below:

This includes Bax, which heterodimerizes with Bcl-2 and **counters** its activity. The **ratio of Bcl-2/Bax** can determine whether a given cell will execute or ignore an apoptotic stimulus (7). (emphasis added, page 821, 1st column).

Given the existence of one regulatory **pair**, Bcl-2 **and** Bax, that an important question arises as to the rationale for further family members. (emphasis added; page 821, 2nd column).

. . .in thymocytes from normal control mice, only 30% of Bax is heterodimerized with Bcl-xL (Fig. 8A, upper panel), while the supernatant of that immunodepletion reveals that 70% of Bax is unbound (Fig. 8A, lower panel). In contrast, in the presence of the Bcl-xL transgene, a substantial portion of Bax (77%) is heterodimerized with Bcl-xL, while only 23% of Bax is unbound (Fig. 8B). The **heterodimerization of >50% of Bax with either Bcl-2 or Bcl-xL resulted in repression of cell death** in a cell line system (20). (emphasis added; page 825, 2nd column to page 821, 1st column).

Chao refers to the importance this parameter (**heterodimerization** of Bcl-2 or of Bax) and the importance of **the ratio of the pair** Bcl-2/Bax compared to the unbound Bcl-2 or Bax as being relevant to whether apoptosis is promoted or repressed in a cell. Based on Chao, the person of ordinary skill would have used **the ratio of the heterodimer pair** Bcl-2/Bax compared to the unbound Bax rather than the comparative levels of Bax and/or Bak expression **alone** to attempt to determine oxaliplatin resistance in a cancer cell.

Unlike Chao, the present invention does not require determination of the ratio of Bcl-2: Bax and there is no suggestion in Chao for the comparative process of the invention. Chao teaches that the inhibition of Bax by heterodimerization with Bcl-xL is associated with an enhanced resistance to oxaliplatin and is silent about whether Bax gene expression alone correlates with oxaliplatin resistance. For example, in the invention a reduced level of Bax expression in a cancer cell compared to a control indicates oxaliplatin resistance. However, based on Chao, a high level of Bax expression in a cancer cell could correlate with oxaliplatin

resistance if the Bcl-xl level leading to (according to Chao) heterodimerization of all the Bax protein and inhibition of apoptosis. Similarly, according to Chao, if the level of expression of Bax is weak and Bcl-xl level expression weaker, then it will remain free Bax (not heterodimerized with Bcl-xl. Chao, page 827, 2nd col. comments on the importance of the pair association between Bax and Bcl0-xl “in normal thymocytes, only a minority of Bax (30%) was heterodimerized with the **modest** levels of Bcl-xl”. Clearly, a weak level of expression of Bax is not necessarily correlated with an enhanced resistance to oxaliplatin. Thus, Chao clearly contemplates a different comparison than that required by the invention. Chao is silent about the relative correlation of Bax and/or Bak expression alone between cancer and control cells with oxaliplatin resistance.

In distinction to the prior art, the invention correlates a reduced expression of a “marker gene expressing the pro-apoptotic Bax and/or Bak protein(s)” compared to a control cell to oxaliplatin resistance.

None of the cited documents suggests that a reduced level of Bax and/or Bak expression in a cancer cell **taken alone in comparison to a control** cell is indicative of oxaliplatin resistance. Therefore, this rejection cannot be sustained.

Issue B: Rejection—35 U.S.C. §103

Claims 1, 2, 5, 8, 10-12, 24 and 27 were rejected under 35 U.S.C. §103(a) as being unpatentable over Maurer, et al., Dig. Dis. Sci. 43:2641, in view of Macpherson, et al., PAACR Ann. Mtg. 43:407 and Chao, et al., J. Exp. Med. 182:821, and further in view of Aggarwal, et al., J. Immunol. 160:1627.

Maurer, et al., Dig. Dis. Sci. 43:2641, Macpherson, et al., PAACR Ann. Mtg. 43:407 and Chao, et al., J. Exp. Med. 182:821 have been discussed above in conjunction with Issue A and do not disclose or suggest the invention.

Aggarwal was applied as teaching a quantitative PCR method, but does not remedy the deficiencies in the other applied references as Aggarwal is not concerned with oxaliplatin resistance. In view of the above, this rejection cannot be sustained.

The arguments above apply at least in equal force to each pending claim. The dependent claims all contain further limitations that establish their patentability apart from those in the independent claims.

RELIEF REQUESTED

The Appellants respectfully request reversal of the grounds of rejection above and the allowance of this application.

Respectfully submitted,

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A handwritten signature in black ink, appearing to read "Thomas Cunningham", written in a cursive style.

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(viii) Claims Appendix

Claim 1 (Previously Presented): A process for detecting resistance of a cancer cell to oxaliplatin treatment comprising:

detecting the expression of an effector or marker gene expressing the pro-apoptotic Bax and/or Bak protein(s) in a cancer cell and in a control cell not resistant to oxaliplatin;
comparing said expression between said cancer cell and said control cell, wherein reduced expression of said effector or marker gene in said cancer cell compared to said control cell indicates that said cancer cell is resistant to oxaliplatin.

Claim 2 (Previously Presented): The process of claim 1, wherein the cancer cell is selected from the group consisting of a cell obtained from a subject having colorectal cancer, a cancer of the ovaries, a cancer of the germinal cells, a cancer of the lung, a cancer of the digestive tract, a cancer of the prostate, a cancer of the pancreas, a cancer of the small intestine, and a cancer of the stomach.

Claim 3-4 (Cancelled)

Claim 5 (Previously Presented): The process of claim 1, comprising detecting mRNA transcripts of said effector or marker gene.

Claim 6 (Withdrawn): The process of claim 1, comprising detecting the amount and/or the activity of at least one mitochondrial apoptosis protein in the cancer cells.

Claim 7 (Withdrawn): A process for *in vitro* detection of the resistance of cancer cells to oxaliplatin treatment comprising:

detecting at least one mutation indicative of deficient mitochondrial apoptosis in the case of treatment with oxaliplatin, in particular of a mutation in a region of the Bax gene containing a series of 8 deoxyguanines.

Claim 8 (Previously Presented): The process according to claim 1 comprising:

- a) determining the level of expression of said effector or marker gene in cancer cells obtained from a patient;
- b) comparing the level measured with the level measured in a corresponding control sample of cells not resistant to oxaliplatin.

Claim 9 (Withdrawn): The process according to claim 6 comprising:

- contacting an antibody that recognizes a mitochondrial apoptosis protein with a sample suspected of containing an apoptosis protein, and
- detecting the formation of an antigen-antibody complex between said antibody and said apoptosis protein;
- wherein a reduced level of complex formation between said antibody and said apoptosis protein compared to the level in a corresponding control cell not resistant to oxaliplatin is indicative of resistance to oxaliplatin.

Claim 10 (Previously Presented): The process of claim 1, wherein a probe or primer is used to detect the expression of said effector or marker gene.

Claim 11 (Previously Presented): The process of claim 1 comprising:

- a) isolating mitochondrial DNA from a biological sample to be examined, or obtaining a cDNA from the RNA of the biological sample or from genomic DNA; and

b) amplifying the DNA from a) using at least one primer for amplification of said effector or marker gene.

Claim 12 (Previously Presented): The process according to claim 1, comprising:

- a) contacting a nucleotide probe for said effector or marker gene with a biological sample to be analyzed for a time and under conditions suitable for hybridization to occur; and
- b) detecting hybridization.

Claim 13 (Withdrawn): A process for selection of compounds that inhibit the resistance of cancer cells to oxaliplatin comprising:

- a) adding at least one candidate compound to the cancer cells resistant to oxaliplatin;
- b) comparing the level of mitochondrial apoptosis and/or expression of at least one apoptosis gene in the presence and absence of the compound;
- c) deducing the anti-resistance effect when the level of mitochondrial apoptosis is greater after addition of the compound, or when the level of expression is greater when the gene is a gene that stimulates mitochondrial apoptosis, or when the level of expression is less when the gene is a gene that inhibits mitochondrial apoptosis.

Claims 14-18 (Cancelled)

Claim 19 (Withdrawn): A kit for diagnosis of resistance of a cancer to oxaliplatin comprising:

- a) at least one compartment suitable to contain a probe;
- b) reagents necessary for the implementation of a hybridization reaction;
- c) at least one primer and the reagents necessary for a DNA amplification reaction.

Claim 20 (Withdrawn): Cell HCT116/S as registered on 16 June 2003, under number: I-3051, with the Collection Nationale de Cultures de Microorganismes (CNCM), Pasteur Institute, Paris, France.

Claim 21 (Withdrawn): A method for using cell HCT116/S according to claim 20, or of any cell derived from this cell HCT116/S, to study the correlation between the resistance of cancer cells, most preferably colorectal, to anti-cancer treatment and the expression of a mitochondrial apoptosis gene.

Claim 22 (Withdrawn): A method of using cell HCT116/S according to claim 20, or of any cell derived from this cell HCT116/S, for the visualization and identification of a mitochondrial apoptosis gene whose expression is linked to the resistance of cancer cells, most preferably colorectal, to anti-cancer treatment.

Claim 23 (Withdrawn): A method for using cell HCT116/S according to claim 20, or of any cell derived from this cell HCT116/S, for the selection of a compound capable of stimulating mitochondrial apoptosis in a cancer cell, said compound being designed to be combined with an anti-cancer agent to which said cancer cell is resistant, most preferably said anti-cancer agent to which said cancer cell is resistant being oxaliplatin and, as the case may be, said cell is a colorectal cancer cell.

Claim 24 (Previously Presented): The process of claim 1, wherein said cancer cell is a colorectal cancer cell, and said detecting comprises detecting the level of expression of mRNA encoding Bax, wherein reduced expression of mRNA

encoding Bax compared to a control cell not resistant to oxaliplatin correlates with resistance of the cancer cell to oxaliplatin.

Claims 25-26 (Cancelled)

Claim 27 (Previously Presented): A process for detecting resistance of a cancer cell to oxaliplatin treatment comprising:

measuring the level of expression of the mRNA(s) encoding the pro-apoptotic Bax and/or Bak protein(s) in a cancer cell and in a control cell not resistant to oxaliplatin;

comparing said expression between the cancer cell and the control cell,

wherein a lower level of expression of said mRNA in the cancer cell compared to a control cell not resistant to oxaliplatin is indicative of resistance to oxaliplatin.

(ix) Evidence Appendix

(none)

(x) Related Proceedings Appendix

(none)